

CLAIMS

1. A method for maintenance of an undifferentiated stem cell, said method comprising exposing a stem cell to a member of the transforming growth factor-beta (TGF β) family of proteins, a member of the fibroblast growth factor (FGF) family of proteins, or nicotinamide (NIC) in an amount sufficient to maintain the cell in an undifferentiated state for a sufficient amount of time to achieve a desired result.
2. The method of claim 1, wherein the method comprises exposing said cell to two or more of a TGF β family member, an FGF family member, and NIC.
3. The method of claim 1, wherein the method comprises exposing said cell to a TGF β family member, an FGF family member, and NIC.
4. The method of claim 1, wherein the TGF β family member is Activin A.
5. The method of claim 1, wherein the FGF family member is keratinocyte growth factor (KGF).
6. The method of claim 1, wherein said exposing results in growth of said cell.
7. The method of claim 1, wherein said exposing is repeated at least one time.
8. The method of claim 1, wherein said stem cell is a mammalian stem cell.
9. The method of claim 1, wherein said stem cell is a human stem cell.

10. The method of claim 1, wherein said stem cell is an embryonic stem cell.

11. The method of claim 1, wherein the desired result comprises culturing said stem cell for ten passages or more.

12. The method of claim 1, wherein the desired result comprises culturing said stem cell for thirty passages or more.

13. The method of claim 1, wherein the TGF β family member shows 30% or greater sequence identity with SEQ ID NO:1.

14. The method of claim 1, wherein the TGF β family member shows 80% or greater sequence identity with SEQ ID NO:1.

15. The method of claim 1, wherein the TGF β family member shows 90% or greater sequence identity with SEQ ID NO:1.

16. The method of claim 1, wherein the TGF β family member shows 95% or greater sequence identity with SEQ ID NO:1.

17. The method of claim 1, wherein the TGF β family member shows 99% or greater sequence identity with SEQ ID NO:1.

18. The method of claim 1, wherein the FGF family member shows 30% or greater sequence identity with SEQ ID NO:17.

19. The method of claim 1, wherein the FGF family member shows 80% or greater sequence identity with SEQ ID NO:17.

20. The method of claim 1, wherein the FGF family member shows 90% or greater sequence identity with SEQ ID NO:17.

21. The method of claim 1, wherein the FGF family member shows 95% or greater sequence identity with SEQ ID NO:17.

22. The method of claim 1, wherein the FGF family member shows 99% or greater sequence identity with SEQ ID NO:17.

23. A composition comprising a) a culture medium and b) a TGF β family member, an FGF family member, NIC, or a combination of two or more of these.

24. The composition of claim 23, wherein the TGF β family member is Activin A.

25. The composition of claim 23, wherein the FGF family member is KGF.

26. The composition of claim 23, further comprising a stem cell.

27. The composition of claim 26, wherein said stem cell is a mammalian stem cell.

28. The composition of claim 26, wherein said stem cell is a human stem cell.

29. The composition of claim 26, wherein said stem cell is an embryonic stem cell.

30. The composition of claim 23, wherein the TGF β family member shows 30% or greater sequence identity with SEQ ID NO:1.

31. The composition of claim 23, wherein the TGF β family member shows 80% or greater sequence identity with SEQ ID NO:1.

32. The composition of claim 23, wherein the TGF β family member shows 90% or greater sequence identity with SEQ ID NO:1.

33. The composition of claim 23, wherein the TGF β family member shows 95% or greater sequence identity with SEQ ID NO:1.

34. The composition of claim 23, wherein the TGF β family member shows 99% or greater sequence identity with SEQ ID NO:1.

35. The composition of claim 23, wherein the FGF family member shows 30% or greater sequence identity with SEQ ID NO:17.

36. The composition of claim 23, wherein the FGF family member shows 80% or greater sequence identity with SEQ ID NO:17.

37. The composition of claim 23, wherein the FGF family member shows 90% or greater sequence identity with SEQ ID NO:17.

38. The composition of claim 23, wherein the FGF family member shows 95% or greater sequence identity with SEQ ID NO:17.

39. The composition of claim 23, wherein the FGF family member shows 99% or greater sequence identity with SEQ ID NO:17.

40. A composition comprising a combination of two or more of a) at least one purified TGF β family member protein, b) at least one purified FGF family member protein, and 3) purified NIC.

41. The composition of claim 40, which is a culture medium for stem cells.
42. The composition of claim 41, wherein the stem cells are mammalian stem cells.
43. The composition of claim 41, wherein the stem cells are human stem cells.
44. The composition of claim 41, wherein the stem cells are embryonic stem cells.
45. The composition of claim 41, wherein the TGF β family member is Activin A.
46. The composition of claim 41, wherein the FGF family member is KGF.
47. An undifferentiated, pluripotent stem cell derived from a culture that has been passaged at least 10 times.
48. The stem cell of claim 47, wherein the culture has been passaged at least 20 times.
49. The stem cell of claim 47, wherein the culture has been passaged at least 30 times.
50. The stem cell of claim 47, wherein the stem cell has been passaged in the presence of media comprising a TGF β family member, an FGF family member, or NIC, but not in the presence of conditioned media, feeder cells, or LIF.

51. The stem cell of claim 47, wherein the stem cell is a mammalian stem cell.
52. The stem cell of claim 47, wherein the stem cell is a human stem cell.
53. The stem cell of claim 47, wherein the stem cell is an embryonic stem cell.
54. A stem cell produced by a method comprising exposing a stem cell to a member of the transforming growth factor-beta (TGF β) family of proteins, a member of the fibroblast growth factor (FGF) family of proteins, or nicotinamide (NIC) in an amount sufficient to maintain the cell in an undifferentiated state .
55. A kit comprising two or more of the following:
- a) a TGF β family member,
 - b) an FGF family member,
 - c) NIC,
 - d) a stem cell, and
 - e) stem cell culture media.
56. The kit of claim 55, wherein the TGF β family member is Activin A.
57. The kit of claim 55, wherein the FGF family member is KGF.
58. The kit of claim 55, wherein the stem cell is a mammalian stem cell.
59. The kit of claim 55, wherein the stem cell is a human stem cell.
60. The kit of claim 55, wherein the stem cell is an embryonic stem cell.

61. A method for maintenance of an undifferentiated stem cell, said method comprising exposing a stem cell to a member of the transforming growth factor-beta (TGF β) family of proteins, a member of the fibroblast growth factor (FGF) family of proteins, or nicotinamide (NIC) in an amount sufficient to maintain the cell in an undifferentiated state for a sufficient amount of time to achieve a desired result, wherein the stem cell is not also exposed to a feeder cell, conditioned media, or leukemia inhibitory factor.

62. A composition comprising a) a culture medium and b) a TGF β family member, an FGF family member, NIC, or a combination of two or more of these, wherein said composition does not comprise feeder cells, conditioned media, or LIF.

63. A differentiated cell derived from a stem cell grown or maintained by a method comprising exposing a stem cell to a member of the transforming growth factor-beta (TGF β) family of proteins, a member of the fibroblast growth factor (FGF) family of proteins, or nicotinamide (NIC) in an amount sufficient to maintain the cell in an undifferentiated state.

64. An embryonic stem cell grown or maintained by a method comprising exposing a stem cell to a member of the transforming growth factor-beta (TGF β) family of proteins, a member of the fibroblast growth factor (FGF) family of proteins, or nicotinamide (NIC) in an amount sufficient to maintain the cell in an undifferentiated state.

65. A pharmaceutical composition comprising a stem cell grown or maintained by a method comprising exposing a stem cell to a member of the transforming growth factor-beta (TGF β) family of proteins, a member of the fibroblast growth factor (FGF) family of proteins, or nicotinamide (NIC) in an amount sufficient to maintain the cell in an undifferentiated state.

66. A cell derived from a stem cell grown or maintained by a method comprising exposing a stem cell to a member of the transforming growth factor-beta (TGF β) family of proteins, a member of the fibroblast growth factor (FGF) family of proteins, or nicotinamide (NIC) in an amount sufficient to maintain the cell in an undifferentiated state